281 Supplemental Figure 1. T. cruzi amastigotes maintain a close association with the host 282 **mitochondria.** Bar graphs representing the % of grouped (A and B) or individual (C and D) 283 intracellular T. cruzi amastigotes that are proximal to (p) or distant (d) from the host Golgi 284 apparatus (A; p value = 0.36) or the MTOC (B; p value =0.013), ER (C) or mitochondria (D; p value <0.0001) accompanied of representative fluorescence images. n > 200 parasites for each 285 of 3 independent experiments. E, F and G. Immunofluorescence images of infected NHDF fixed 286 287 at 48hpi stained with ATP5B antibodies (green) (E) or TOM20 antibodies (green) (F,G). 288 Parasites were stained with rabbit anti-T. cruzi antiserum (red) and DNA was stained with DAPI 289 (blue). Scale bar = $5 \mu m$.

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291	Supplemental Movies 1 and 2. Fluorescence time-lapse movie of adherent NHDF expressing
292	mCherry in the mitochondrial matrix (mito-mcherry, red) and infected with GFP-expressing T.
293	cruzi parasites (green) (36 hpi) showing the proximity between the T. cruzi amastigotes and the
294	host mitochondria. Images were taken every 5 min and the movie is designed to play 3 frames
295	per second. Time of recording is indicated in the upper left corner of each frame.

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Supplemental Movies 3 and 4. Fluorescence time-lapse movie of *T. cruzi*-GFP infected NHDF-mito-mCherry (36 hpi) initiated 180 min post-replating following trypsinization, highlight the dynamics of the host mitochondrial network in live cells (red) and the proximity between host mitochondria and intracellular amastigotes (green) during cell spreading. Sample was imaged every 5 min and the movie is designed to play 3 frames per second. Time of recording is indicated in the upper left corner of each frame.

Supplemental Figure 1



224x147mm (300 x 300 DPI)